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Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

On

February 1, 2005

TOWNSEND and TOWNSEND and CREW LLP

By

Raven Karlin

PATENT
Attorney Docket No.: 018512-006810US

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:

JEGLA, Timothy J.

Application No.: 09/921,159

Filed: August 1, 2001

For: SLO2 AND SLO4, NOVEL
POTASSIUM CHANNEL PROTEINS
FROM HUMAN BRAIN

Customer No.: 20350

Confirmation No. 7080

Examiner: PAK, Michael D.

Technology Center/Art Unit: 1646

DECLARATION UNDER 37 C.F.R. §1.132
OF DR. MCCORMACK

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

I, Ken McCormack, being duly warned that willful false statements and the like are punishable by fine or imprisonment or both (18 U.S.C. § 1001), and may jeopardize the validity of the patent application or any patent issuing thereon, state and declare as follows:

1. All statements herein made of my own knowledge are true, and statements made on information or belief are believed to be true and correct.

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Declaration under 37 CFR 1.132 of Dr. McCormack

2. I received a B.A. degree in Psychobiology from the University of California at Santa Cruz in 1985 and a Ph.D. in Neurobiology from the California Institute of Technology in 1991. I served as a postdoctoral fellow in the Department of Cellular and Molecular Biology at Yale University from 1991-92, the Max Planck Institute for Biophysical Chemistry and Experimental Medicine from 1992-95 and the Department of Genetics, Pathobiology and Physiology at the University of Wisconsin at Madison from 1995-97. I was the Group Leader for Molecular Biology at Arcaris/Deltagen Proteomics from 1997-2000 and a Principal Scientist at Aurora Biosciences/Vertex Pharmaceuticals from 2000-04. Currently, I am the Group Leader for Molecular Biology at Icagen, Inc., and have been at this position for 6 months. A copy of my curriculum vitae is attached as Exhibit A.

3. The invention of the above-referenced patent application provides for the first time nucleic acids encoding human Slo2 and Slo4, novel members of the Slo family of voltage-gated potassium channels that is highly expressed in the central nervous system (CNS) and in some peripheral tissue at lower levels.

4. I have read and am familiar with the contents of this patent application. In addition, I have read the Office Action, mailed October 6, 2004, received in the present case. It is my understanding that the Examiner asserts that the present invention is not supported by a specific, substantial, and credible asserted utility or a well established utility as required by the United States patent laws.

5. This declaration is provided to demonstrate that the identification of the coding sequences for human Slo2 and Slo4 has a specific and substantial utility that is credible to one of ordinary skill in the art.

6. The Slo2 and Slo4 channels are voltage-gated potassium channels highly expressed in the central nervous system (CNS). Since these potassium channels begin to activate in a voltage range below the typical thresholds for action potential generation, one of skill in the art would reasonably believe that the channels are involved in modulating cell excitability. Because of their high level of expression in the CNS, an artisan would also reasonably believe

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Declaration under 37 CFR 1.132 of Dr. McCormack

that these channels can serve as therapeutic targets for treatment of conditions related to altered neuronal excitability in the CNS, *e.g.*, epilepsy, migraines, and psychotic disorders. The identification of the Slo2 and Slo4 channels therefore has a substantial utility, or a "real world" use, since this discovery makes possible the routine identification of activators and inhibitors of the Slo2/Slo4 channels, which may be used as therapeutic agents for treating conditions caused by or related to abnormalities in neuronal excitability. This utility relies on the expression of Slo2/Slo4 channels in the CNS and their involvement in the regulation of cell excitability, which are specific features of these potassium channels and not a broad class of ion channels. The present invention thus has a specific utility.

7. The present invention not only provides nucleic acids encoding human Slo2 and Slo4, but also teaches methods for detecting the activity of the Slo2/Slo4 channels (*see, e.g.*, page 49, line 7, to page 52, line 29; and page 74, lines 7-21, of the specification) and methods for identifying modulators of the ion channels (*see, e.g.*, page 52, line 32, to page 57, line 12). Upon reading this disclosure, a skilled artisan would be able to readily screen candidate compounds and identify activators or inhibitors of a Slo2/Slo4 channel, without the need to carry out extensive additional research. The present invention therefore has a real-world use.

8. There are known instances where modulation of an ion channel is useful for treating a specific disease even though the ion channel itself may not cause the disease. For example, hypertension can be caused by a variety of illnesses such as renal disease and diabetes. Among the treatment strategies for hypertension is the use of drugs such as calcium channel blockers to relax the vasculature. Relaxing the vasculature to reduce blood pressure is useful and effective, even if the original cause of the hypertension is unrelated to vascular tone. Similarly, it is perfectly reasonable to expect that the targeting of a Slo2/Slo4 potassium channel, a voltage-gated potassium channel highly expressed in the CNS and involved in regulation of neuronal cell excitability, is an appropriate strategy for treating neurological disorders related to abnormal excitability of the cells, whether or not such abnormality is directly caused by altered Slo2/Slo4 channel activity. Thus, the asserted utility of the Slo2/Slo4 potassium channel of the present application is reasonable and therefore credible to an ordinarily skilled artisan.

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Declaration under 37 CFR 1.132 of Dr. McCormack

9. In summary, it is my scientific opinion that one of skill in the art, at the time the application was filed, would believe the physiological role human Slo2 or Slo4 plays in the modulation of cell excitability in the CNS and recognize the specific and real-world utility of the Slo2 or Slo4 encoding nucleic acids of the present invention.

Date: January 5, 2005By: Ken McCormack

Ken McCormack, Ph.D.

TOWNSEND and TOWNSEND and CREW LLP
Two Embarcadero Center, Eighth Floor
San Francisco, California 94111-3834
Tel: 415-576-0200
Fax: 415-576-0300
Attachment (Exhibit A: Dr. McCormack's CV)
CG/cg
60371762 v1

Ken McCormack, PhD

ICAgen, Inc.
4222 Emperor Blvd Suite 460
Durham, NC 27703
919-941-5206 ext. 539
kmccormack@icagen.com

Home: 11317 John Allen Rd
Raleigh, NC 27614

Industrial Experience

ICAgen Inc.

Group Leader

6/01/04-present

Team leader for the molecular biology group. Responsibilities include generation of molecular reagents including cDNAs, cell lines, etc. for ion channel drug discovery efforts and evaluation of new targets through RNA expression and target validation strategies including molecular (e.g., siRNA) and pharmacological approaches.

Vertex/Aurora Biosciences, San Diego CA

Principal Scientist

6/15/2000-5/15/04

Vertex: Ligand and voltage-gated ion channel drug discovery project development. Evaluate, present and coordinate early-stage discovery efforts including target selection. Initiated discovery program for gene family with inflammatory and neuropathic pain indications. Biology team leader/coordinator for early-stage discovery effort (target/assay/HTS/hit validation/med chem support/secondary assay development) targeting neuropathic pain. Head of molecular biology for ion channel group. Provide primary scientific support for several business development efforts.

Aurora: Assay development and HTS for ion channel targets and transporters using fluorescent Ca⁺⁺ or voltage-dependent dyes or halide-sensitive YFP (1). HTS and UHTSS screening and hit validation. Project leader and liaison for several external collaborative projects. Profiling of endogenous targets in primary cells; generation of immortalized primary cells including neurons.

Deltagen Proteomics/Arcaris Inc., Salt Lake UT Group Leader

8/97-5/2000

Oncogenic assay development; immortalization of human primary cells and tumor formation in nude mice with limited sets of oncogenes. Utilization of yeast two-hybrid (Y2H) peptide-binders (to HPV oncogenes) for disruption of protein-protein interactions and development of peptide-target small molecule displacement screening. Received phase I SBIR for screening peptide-inhibitors of HPV E6 and E7 proteins.

Established in-house mammalian cell culture, cDNA libraries and expression systems (retroviral). Design and development of large-scale phenotypic selections (cell cycle arrest, growth factor dependence, apoptosis) and screening of peptide and cDNA expression libraries. Generation of neuronal cAMP-responsive GFP transcriptional reporter cell lines for identification of CRE-regulatory "perturbagens". Responsible for project coordination with academic collaborators (NIH grant).

Academic Research

NIH Postdoctoral Research Fellow

UW Madison

2/95-7/97

Functional characterization of two distinct K⁺ channel associated β subunit genes; generation of null-mutant mice for (Kv β 1 and Kv β 2) and a "knock-in" point mutation of Kv β 2 (2). Gene mapping/cloning,

vector construction, ES cell culture, mouse breeding, phenotyping and development of PCR-based genotyping. GST-fusion purification of Kv β proteins.

HFSP Postdoctoral Fellow

Max-Planck-Institute, Goettingen, Germany

12/92-1/95

Cloning and characterization of the human Kv β 1 and Kv β 2 genes, including the first functional expression of the major Kv1 family β subunit, Kv β 2 (5), chromosomal mapping (4), analyses of splice products, structural modeling based on "data-mining" and limited homologies with NAD(P)H-dependent oxidoreductases (6) (subsequently verified by X-ray crystallographic studies). Pharmacological, molecular, electrophysiological and kinetic modeling analyses of voltage-gated K⁺ channel (α subunit) conformational activation. Mechanistic characterization of state-dependent drug-channel interaction using single-channel, gating current and whole cell K⁺ conductance properties in conjunction with mutagenized differentially-4-AP-sensitive channel constructs (7).

NIH Postdoctoral Assistant

Yale University, New Haven, CT

4/91-10/92

Site-directed mutagenesis, electrophysiological analyses and the first quantification of the voltage-dependent conformational changes of the integral membrane Shaker K⁺ channel proteins. Analyses of voltage-dependence in gating-mutant channels and multisubunit constructs (8-10).

Education:

1991 Ph.D.,	California Institute of Technology	<i>Molecular/Neurobiology</i>
1985 B.S.	UC Santa Cruz	<i>Neurobiology</i>

PhD student

CalTech, Pasadena, CA

1985-1991

Site-directed mutagenesis and initial electrophysiological analyses of "leucine zipper" motif gating-mutants in Shaker K⁺ channels. Utilization of gating-mutants and distinct Shaker splice products to determine multimeric nature of K⁺ channels. Chromosomal walking, library screening to determine unique splice products (11-13).

Bachelor of Science

UC Santa Cruz, CA

1983-1985

Electrophysiological and pharmacological characterization of muscarinic response involved in a molluscan behavioral response (14).

Awards, Fellowships and Memberships

Medicinal Chemistry series UCSD Extension	6/2002
SBIR National Cancer Institute, Arcaris/Deltagen Proteomics	4/2000
NIH Postdoctoral Fellowship National Institute of Health, UW Madison	95-97
Foreign Postdoctoral Fellow Max Planck Society, MPI fur experimentelle Medizin	94-95
Postdoctoral Fellow Human Frontiers Science Program, MPI fur biophysikalische Chemie	92-93
Member Society for Neuroscience	1992- present

Technical Expertise

Including but not limited to:

Molecular Biology; generation of genomic and cDNA libraries, RNA isolation, cDNA synthesis, library screening, RT-PCR, Northern blots, site-directed PCR and cassette mutagenesis, quantitative PCR coupled

with FACS and phenotypic enrichment cycling, isolation of novel cDNAs from degenerate PCR, generation of transcriptionally active reporters.

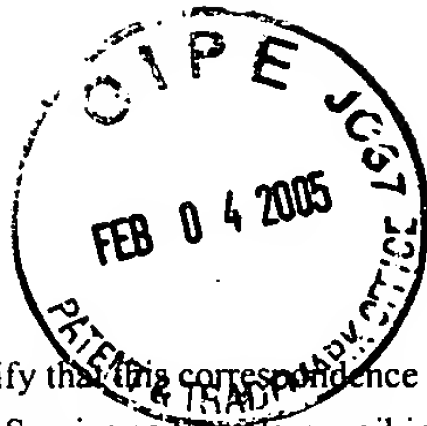
Cell Biology and Biochemistry; Assay development (fluorescent optical readout or phenotypic - cell cycle arrest, apoptosis, etc.), drug discovery HTS and med chem support, pharmacological characterization of ion channels and GPCRs, cell culture including primary and ES cells, transfection and retroviral transduction, immortalization of human primary cells, yeast two hybrid (Y2H) screening, Western blotting, immunofluorescence, monoclonal antibody purification, affinity, DEAE, size-exclusion and FPLC chromatography, protein expression/isolation from *E. Coli*, *Xenopus* oocytes and mammalian tissue.

Biophysical & Computational; patch (including single-channel and gating current recordings) and two-electrode recordings, FACS sorting and analysis, bioinformatic "data-mining", structural and kinetic modeling.

Publications

1. McCormack, K., Heim, R., Raj, D., Xu, J. and Gonzalez, J. Mutant yellow fluorescent protein (YFP) (H148Q) as a cell-based probe for HTS of GABA receptors . In preparation. (Neuroscience 2001 abstract #911.11)
2. McCormack K, Connor JX, Zhou L, Ho LL, Ganetzky B, Chiu SY, Messing A.. Genetic analysis of the mammalian K⁺ channel β subunit Kv β 2 (KCNA2). *J Biol Chem*. 2002 277(15):13219-28.
3. McCormack T, McCormack K, Nadal MS, Vieira E, Ozaita A, Rudy B. (1999) The effects of Shaker beta-subunits on the human lymphocyte K⁺ channel Kv1.3. *J Biol Chem*, 274(29):20123-6.
4. Schultz D, Litt M, Smith L, Thayer M, McCormack K. (1996). Localization of two potassium channel beta subunit genes, KCNA1B and KCNA2B. *Genomics* 31(3):389-91.
5. McCormack K, McCormack T, Tanouye M, Rudy B, Stuhmer W. (1995). Alternative splicing of the human Shaker K⁺ channel beta 1 gene and functional expression of the beta 2 gene product. *FEBS Lett*. 370(1-2):32-6.
6. McCormack, T. and McCormack K. (1994). Shaker K⁺ channel beta subunits belong to an NAD(P)H-dependent oxidoreductase superfamily. *Cell* 79(7):1133-5.
7. McCormack K, Joiner WJ, Heinemann SH. (1994). A characterization of the activating structural rearrangements in voltage-dependent Shaker K⁺ channels. *Neuron* 12(2):301-15.
8. McCormack K, Lin L, Sigworth FJ. (1993). Substitution of a hydrophobic residue alters the conformational stability of Shaker K⁺ channels during gating and assembly. *Biophys J*. 65(4):1740-8.
9. Schoppa NE, McCormack K, Tanouye MA, Sigworth FJ. (1992). The size of gating charge in wild-type and mutant Shaker potassium channels. *Science* 255(5052):1712-5.
10. McCormack K, Lin L, Iverson LE, Tanouye MA, Sigworth FJ. (1992). Tandem linkage of Shaker K⁺ channel subunits does not ensure the stoichiometry of expressed channels. *Biophys J*. 63(5):1406-11.
11. McCormack K, Tanouye MA, Iverson LE, Lin JW, Ramaswami M, McCormack T, Campanelli JT, Mathew MK, Rudy B. (1991). A role for hydrophobic residues in the voltage-dependent gating of Shaker K⁺ channels. *Proc Natl Acad Sci U S A* 88(7):2931-5.

12. McCormack K, Lin JW, Iverson, LE, Rudy B. (1990). Shaker K⁺ channel subunits from heteromultimeric channels with novel functional properties. **Biochem Biophys Res Commun.** 171(3):1361-71.
13. McCormack, K., Campanelli, J.T., Ramaswami, M., Mathew M., Tanouye, M.A. (1989). Leucine zipper motif update. **Nature** 340, 103-104.
14. Morielli AD, Matera EM, Kovac MP, Shrum RG, McCormack KJ, Davis WJ. (1986). Cholinergic suppression: a postsynaptic mechanism of long-term associative learning. **Proc Natl Acad Sci U S A.** 83(12):4556-60.



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PATENT
Attorney Docket No.: 018512-006810US

Commissioner for Patents
P.O. Box 1450
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On February 1, 2005

TOWNSEND and TOWNSEND and CREW LLP

By: Karen Karlin

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:

JEGLA, Timothy J. et al.

Application No.: 09/921,159

Filed: August 1, 2001

For: SLO2 AND SLO4, NOVEL
POTASSIUM CHANNEL PROTEINS
FROM HUMAN BRAIN

Customer No.: 20350

Confirmation No. 7080

Examiner: Michael D. Pak

Technology Center/Art Unit: 1646

DECLARATION OF TIMOTHY JAMES
JEGLA AND JULIE DICKSON WITZEL
PURSUANT TO 37 C. F. R. §1.131

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

We, Timothy James Jegla and Julie Dickson Witzel, being duly warned that willful false statements and the like are punishable by fine or imprisonment or both, under 18 U.S.C. §1001, and may jeopardize the validity of the patent application or any patent issuing thereon, state and declare as follows:

1. All statements herein made of our own knowledge are true and statements made on information or belief are believed to be true. Exhibit I is attached hereto and is incorporated herein by reference.

2. At the time this invention was first conceived, we were employees of ICAGEN, Inc., located in Durham, North Carolina. All activities described in this Declaration took place in the United States of America.

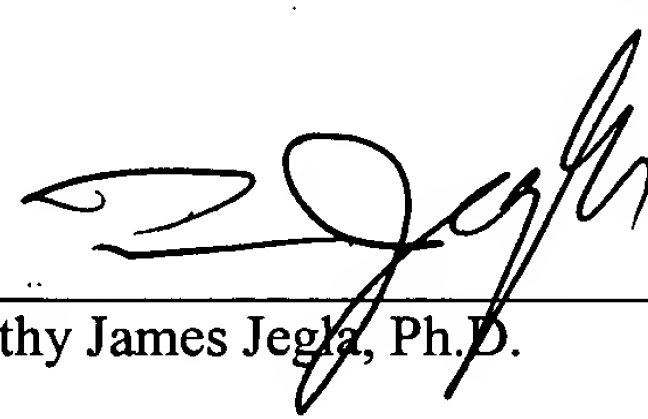
3. In accordance with 37 C.F.R. §1.131, we state that we conceived the claimed invention and reduced it to practice in the United States prior to July 31, 2000, which is the earliest possible priority date of the Curtis *et al.* reference (U.S. patent application published as US 2003/0143675) for SEQ ID NO:58 disclosed therein.

4. Attached to this Declaration is Exhibit I, which contains copies of laboratory notebook entries demonstrating the possession of SEQ ID NO:2 and SEQ ID NO:4 of the present invention. The dates in the Exhibit have been redacted. All redacted dates are prior to July 31, 2000.

5. Conception of the present invention as well as its reduction to practice are evidenced by Exhibit I. On pages 88-89 of Exhibit I, the polynucleotide coding sequence and corresponding amino acid sequence for human Slo4 (SEQ ID NOs:3 and 4, respectively) are provided. On pages 125-127, the polynucleotide coding sequence and corresponding amino acid sequence for human Slo2 (SEQ ID NOs:1 and 2, respectively) are provided.

6. In light of the foregoing, it is established that Declarants had in their possession the claimed subject matter of the present invention prior to July 31, 2000. As such, the Curtis *et al.* reference is antedated, even assuming that the reference is entitled to the July 31, 2000, priority date for SEQ ID NO:58 disclosed therein.

Dated: 1/12/05

By: 
Timothy James Jegla, Ph.D.

Dated: _____

By: _____
Julie Dickson Witzel, Ph.D.

Attachments (Exhibit I: redacted copies of pages 88-89 and 125-127 of laboratory notebook)
60381926 V1



Appl. No. 09/921,158
Declaration under 37 C.F.R. §1.131
Reply to Office Action of October 6, 2004

PATENT

2. At the time this invention was first conceived, we were employees of ICAGEN, Inc., located in Durham, North Carolina. All activities described in this Declaration took place in the United States of America.

3. In accordance with 37 C.F.R. §1.131, we state that we conceived the claimed invention and reduced it to practice in the United States prior to July 31, 2000, which is the earliest possible priority date of the Curtis *et al.* reference (U.S. patent application published as US 2003/0143675) for SEQ ID NO:58 disclosed therein.

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Dated: _____

By: _____
Timothy James Jegla, Ph.D.

Dated: 11/9/05

By: Julie Dickson Witzel
Julie Dickson Witzel, Ph.D.

Attachments (Exhibit I: redacted copies of pages 88-89 and 125-127 of laboratory notebook)
60381926 V1

Map of Slc4. Consensus coding sequence.

ATGTTTGAATTTGGACAGCGAAGTCCCCCTCTGCTCCACGTTACAGGTTTCGACATTTGCTGCTAGGGGACCAAGGATGCCAAAACGACGACAGGCTACAAGTTGAATTCATATGAATGAAAATACATTTA
TACCAACTAAACCTCTCGCTTCACGGGGACAGCGAGGTTCCATGTCCAAAGCTCTAAACGACGATCCCCGGTCTCTACCGTTTCTGCTGCTCCCATGTTCAACTTAAGATATACCTTACTTTTATGTAAT
H V D L E S E V P P L P P R V R F R D L L L G D O G W O N O D R V Q V E F Y H N E N T F

EcoRI
PstI

133

AAGAAACATAAATTTATTTTCATAAAAAACACAGATCAAGTCTAAGGATACGCTGTTCATTTTCTCTCAAATTAAGCTGCTTATTATACATAATCCGAGTACTACTAGAAAACCTTCAACAAG
TTCTTCTGATTTTAATAAAAAAGTATTTTGGTCTCTAGTTTCAGATTCCATGCGGACAAAGTTAAACACAGCTTTAATGATTGACGAATAATATGATTAGGCTCATGATGATCTTTTGGGAAGTGTCC
K E R L K L F F I K N O R S S L R I R L F N F S L K L L S C L L Y I R V L L E N P S O G

SacI

266

AAATGAATGGTCTCATATCTTTTGGTGAACAGAAGTCTACCTTTGTCGGGCTTACAGGTTTCACTGGCATTCATAAGCTCTGTTTGAACAATATTACTTGGTTATCTTAGTTATAAGCGAAACATCTGGGA
TTTACTTACCAGATATAGAAAACCACTTGTCTTCAGATGGAACACCCGAAATGTCAAAGTCAACGTAACATTACAGACAACTTTGTTAATAAGAACCAATAGAATCAATATTCCTTTTGTAGACCTTT
N E W S H I F W V N R S L P L V G L O V S V A L I S L F E T I L L G Y L S Y K E N I W E

SspI

399

CAGATTTACGAATACCTTCATCTTTTGGTGAACAGAAGTCTACCTTTGTCGGGCTTACAGGTTTCACTGGCATTCATAAGCTCTGTTTGAACAATATTACTTGGTTATCTTAGTTATAAGCGAAACATCTGGGA
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O I L R I P F I L E I I N A V P F I I S I F W P S L R N L F V P V F L N C V L A K H A L

VspI SspI XcmI

532

AAATATGATTAATGATCTACACAGAGCCATTACGCTACACAGCTCTGAATGTTTAAATCAAGTTTGATTTTAAATATCTACATTACTATGCTTATCTTACCTGCAATTTGTTGGGATCCAACATCTGGAAG
TTTATCTAATTAAGTATGATGCTCTCGGTAACTGCGATGCTGTCAGACCTTACAAATAGTTTCAAACTAAATATATAGATGTAATGATACGGAATAGAAGTGGAGCTAAACACCTAGGTTGTAGACCTTGC
E N M I N D L H R A I O R T O S A M F N O V L I L I S T L L C L I F T C I C G I O H L E R

VspI BamHI

665

AATAGCAAAGAAGCTGAATCTCTTGAATCTCTTGAATCTGTCAGCTTTTCTACTGTCGGCTTGGGGATGTCACCTCTGAACATGCTCTCTCAAGCTTTTGTAGTTGCTATGATTGCTGTCT
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I G K K L N L F D S L Y F C I V T F S T V C F G D Y T P E T W S S K L F V V A M I C V A

HindIII

799

CTTGTGCTTCTACCCATACAGTTTGAACAGCTGGCTTATTTTGTGATGGAGACAAAAGTCAGGAGGAAACTATAGTGCACATACAGCTCAAACTCAAAAGCATGTCGCTCTGTGTCAGCTCACTGAAGA
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L V V L P I O F E O L A Y L W M E R O K S G G N T S R H R A D T E K H V V L C V S S L K

PvuII SstI SacI SstI

931

TTGATTTACTTATGATTTTAAATGAATTTCTATGCTCATCTAGGCTCCAGGATTTATGTCGGTGAATTTGCTGCTCTACTGAAATGGATGTACAGGTTTCAAGGGTACTGCAGATTCGAATGCTGCTCCCA
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I D L L N D F L N E F T A H P R L O D T Y V V I L C P I E M D Y O V R R V L O I P H W S O

PstI PstI

1064

ACGAGTTATCTACCTTCAAGGTTCAAGCTTAAAGATCAAGACCTATTGAGAGCAAGATGGATGACGCTGAGGCTGTTTATTTCACTAGGCTTGTGAAGTGGATAGGACATCATCTGATCAACCAACA
TGTCTAATAGATGCAAGTTTCAAGCTCGGAATTTCTAGTTCTGGAATACTCTGCTTCTACCTTCTGCGACTTCCGACAAAATAAGAGTATCGGCAACACTTCACTTCTCTGATGACCTAGTGGTTTGT
R V I Y L O G S A L K D O O L L R A K M D D A E A C F I L S S R C E V D R T S S D H O I

SstI PstI

1197

ATTTTACAGCATGGGCTGTGAAGATTTTGTCCAAATTTGCTTTTGTATGTCAGATATTAAGGCTGAAAATAAATTTACATCAAAATTTGCTGATCATGTTGTTTGTGAAGAGGTTTAAATACGCCA
TAAACTCTGTTACCGACACTTTTAAACAGAGGTTTAAACGAAACATACAGGCTCTATAATTTGGACCTTTTATTTAAAGTGTAGTTTAAAGGACTAGTACAAACAACTTCTTCTCAAAATTTATGCGGT
I L R A V A V K D F A P N C P L Y V O I L K P E N K F H I K F A D H V V C E E E F K Y A

PstI XbaI BstXI

1330

TGTACCTTTAAACTGTATATGCCAGCAACATCTACACTTATTACACTACTGTTTATACCTCTAGAGGGCAAGAAGGCCAGCAATGCCAGAACATGGCAGAAGATGTACGCTAGATGCTCCGGGAATGA
ACAATCGAAATTTACATATACGGGCTGTTGATGATGCAATAATGTCATGACCAAGTATGAGATCTCCGCTTCTTCCGGTCTGTACGGGCTTGTACCTGCTTCTACATGCCATCTACGAGGCCCTTACT
H L A L N C I C P A T S T L I T L L V H T S R G O E G O O S P E Q W O K M Y G R C S G N E

PstI XbaI BstXI

1463

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DATE _____

WITNESS

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SCIENTIFIC BINDERY PRODUCTIONS CHICAGO 60605 Made In USA

Work continued to Page

SIGNATURE

DATE

DISCLOSED TO AND UNDERSTOOD BY

DA*

WITNESS

DA

Map of

Slb2 EV

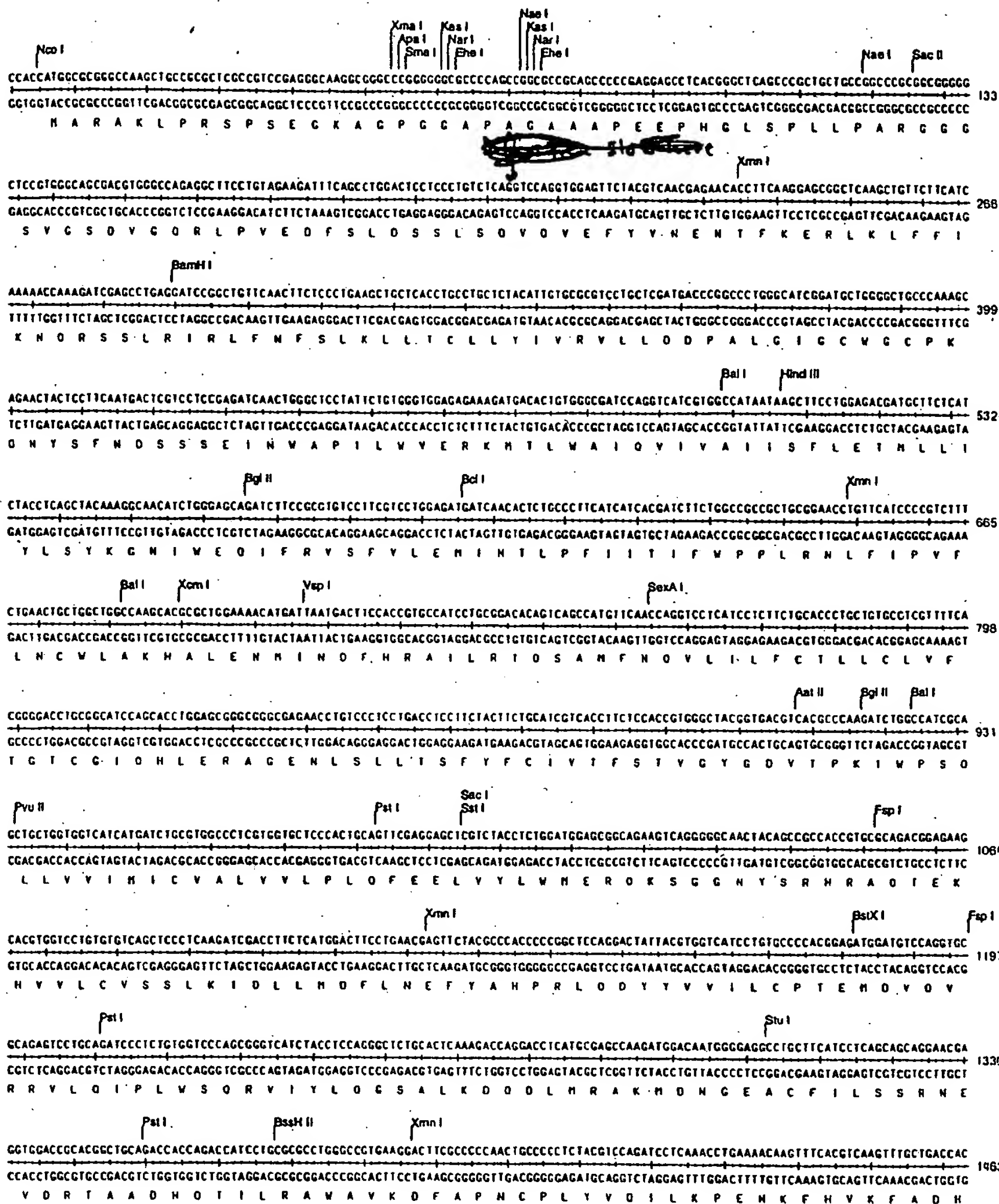
5 Drop insert with
EcoRI/KbaI

& Also Moved
To pCONA3.1
for mammalian
cell expression.

10 (Pgs. 76 & 77)

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BEST AVAILABLE COPY

These clones were the original ones used to define most of the Slb2
Seq. Info before the genomic clone was available. See pages 142-162 for production
of these clones. Pg. 173, 198-202 in book 78 pg. 1-8 in book 101,
for production of the sequences. (3/93)
RAE/ERS that generated original sequences are in books 59, 61 & 68.

[Signature]

[Signature]

KpnI BamHI XmaIII
CCTGGGCTGGACACACCGCGGCTGGGGTACCTCTGTGCGCATGAAATCACCAGGCGGACCTGTGGATCCGCACTACGGCGGCTCTTCCAGAAGCTCTGCTCTCCAGCGCGAGATCCCATTTGGC 3059
CACCCTGGACCTGTGGTGGCGCCGAGCGCCATGGACACACGCTACTTTAGTGGCTCCCGCTGGACACCTAGGCTGATGGCGCGGAGAAGCTCTTCCAGACGAGGAGGTCCGGCTCTAGGGTAACCG
LCLDTTPGSGYLCAKITEGDLVIRTYGRLFORKLCSSSAEIPIG

PstII ApaI
ATACCGGACACAGAGCCAGCTCTTCTCCACCCTGGAGCGCCACGACCTCAGAGCCAGTCCAGATCTGGTGAAGCTGGAGGACTGTGAGGACACACGGGAAGTGAAGGGGCTCCGGCTCCCGGCTG 3192
TAGATGGCTGTCTCTGGTGCAGAAGGTCAGCGCTCCGGTCTGGAGTCTCGGGTACGGTCTAGAGCCACTTGCACCTCTCTGACACTCTGTGTGCTTCACTTCCCGGAGCCCGAGCGCGGAC
IYRTESHVFTSTSEPHOLRAOSOSISVNVEDCEDTREVKGPGVGSRA

PstI ApaI XbaI NotI XmaIII
CCACCGGAGGACGCTCCAGGGCGGCGACACGGCGGCGGTGACCGCGCAGAGCAGCCACTGCTACGGCGAAGAGCTTGCAGTGGCGCGGAGGCTGAGCGCAAGCGCCCAAGCAGCAGCGCGCGGCG 3325
CCTGGCTCTCGTCCAGGCTCCCGCGGTGTGGCGCGGCTCTGGGCTGAGGCTGCGCGCTCTCGGAGTCAACCGCGCTCCGAGTGGCGGTCCCGCGGTTCGCGCGGTTCGTCGCTCCCGCGGCG
CTCGSSSGCRHTGGGCDPAEHPLLRKSLQVARRRLSRKAPKQAGRAA

SacII SacI
GGCGCGGAGTGGATCAGCCAGCAGCGCTCAGCTGTACCGCGCTCTGAGCGCCAGGAGCTCTCCGAGCTGGTGAAGAACCGCATGAAGCAGCTGGGGCTGCCACCCAGCGCTACGAGCAGTAGCAAT 3458
CCCGCGCTCAGCTAGTCCGTCTCGCGGAGTCCGACATGGCGCGGAGCTCGGGTCTCGAGAGCTCGACCACTCTTGGCGTACTTCTGGACCCCGAGCGGTGGTGGCGGATGCTCTGCATCGTTA
AAEVISSOORLSLYRRSEROELSELVKNRHKHLGLPTITGYEDVAN

PstI PvuII PstI NruI XbaI
TTACAGCCAGTGTATCATGAATCGGCTAAACCTGGCATATTTCAGAGCAGATGAAGCAGCAGCAGCAACACCTCTCTAGCTCTCATCAACCTCCCGCGGACAGGCTGGAGCGCAGTGCATTC 3591
AATTGTCGCTCAGTACAGTACTTACCCATTTGGACCTATAACGTTCTGCTACTTCTGCTGGTGTCTTCTGGCAGCAGTGCAGGAGTACTTGGAGCGCGGCTGTGCTCCGAGCTCCGGTCACTGTAA
LTAASDVHNRVNLGYLODEHNDHONTLSYVLIINPPPTDTRLEPSDI

PstI PvuII PstI NruI XbaI
TCTATCTATCCGCTCCGACCCCTGGCTCAGCTGGCCAGCAGCTCCAGAGCGGGAAGCAGCTGCGAGCCACAGCTGTCTCTCTGCAACCCCGAGACTCCGAGCAGACACAGCTCTAATCTAGACCTG 3729
AGATAGATAGCGGAGCTCGCGGAGCGAGTGCACCGCTCTGAGGCTCTCGGCTCTCTGTCAGCTCGGCTGTTCGAGCAGTGTCTGTGTCGAGATAGATCTGGGAC
VYLIRSDPLAHVA'SSSDSRKSSCSHKLSSCNPETRDETOL

Continued Slo2 Map

Partial Slo2 Clone KIAA1422 deposited on
Public Database on 3-14-00
Map on Next 3 pages.

Alignment to Slo2 on Pg. 131

KIAA1422 is 3' Incomplete; 3' End cloning is not obvious
from this clone and was done much
earlier by us.

Also: 5' End Differs (see pg. 131)

2 possible Methionines (A₁₂₂), but ORF not closed
upstream; May be 5' incomplete.

Work continued to Page _____

DATE

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